

Creating a Standardized Approach for Developing Medical Grade 2D Materials

Nichols GP

Nanotechnology Studies Program, ORAU, USA

Corresponding author:

Nichols GP, Program Manager, Nanotechnology Studies Program, ORAU, PO Box 117, MS-23, Oak Ridge, TN, USA, 37831, Tel: (865) 576-3144, Email: Gregory.Nichols@orau.org

Copyright: © 2016 Nichols GP. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Citation: Nichols GP (2016) Creating a Standardized Approach for Developing Medical Grade 2D Materials. Trends in Nanotechnology & Material Science 1: 1-3.

Received Date: March 05-2016

Accepted Date: March 28, 2016

Published Date: April 04, 2016

Abstract

Since the discovery of graphene in 2004, the world of two-dimensional (2D) nanomaterials has rapidly expanded to include a variety of different types. Not only are there multiple types of graphene-related materials, but there are transition metal dichalcogenides, hexagonal boron nitride, and many others. These nanomaterials are showing great promise for biomedical applications because of their large surface area and high mechanical strength. However, any nanomaterial used in medical practice must show that it is biocompatible and has minimal toxic effects. The toxicological data that are usually fairly rigorous regarding most materials is severely lacking among 2D nanomaterials because of their relative newness. Even though toxicological data are limited, there appears to be enough information to notice trends and patterns among some of the properties in the existing 2D nanomaterials that could provide the beginning of a model for standardizing the definition of “medical grade” 2D nanomaterials. Properties such as thickness, surface charge, surface chemistry, and particle size have a large impact on the toxicity of 2D nanomaterials. The literature also shows that some 2D nanomaterials would be more preferred over others for use in biomedical applications, thus a hierarchy of use can begin to be pieced together. As new data on toxicity and physicochemical properties are collected, this model can be updated.

Keywords: Nanotechnology, 2D nanomaterials, Biomedical applications, Safe design

Abbreviations: 2D: Two Dimensional; GFNs: Graphene Family Nanomaterials; ROS: Reactive Oxygen Species; GO: Graphene Oxide; rGO: reduced Graphene Oxide; Cl: Chlorine; Br: Bromine; I: Iodine; TMDs: Transition Metal Dichalcogenides; Mo: Molybdenum; W: Tungsten; Re: Rhenium; S: Sulfur; Se: Selenium; hBN: hexagonal Boron Nitride; B: Boron; N: Nitrogen

Introduction

Two dimensional (2D) nanomaterials have recently become a new phenomenon in nanotechnology. Since the discovery of graphene in 2004, an entirely new universe of similar materials has been developed and continues to emerge on the scene at a blistering pace. These nanomaterials are the thinnest materials known to exist and possess the highest specific surface areas of all nanomaterial types [1]. Because of this property, they have become invaluable for applications requiring high levels of surface interactions on a small scale [1], such as in medical applications where highly complex biological interactions occur. It appears that most 2D nanomaterials do have valuable properties for biomedical applications; however, they are so new that limited data exist, especially toxicological data, which proves difficult when making decisions regarding their safe application in clinical settings and for patient use. In addition, there are a variety of types with a range of toxicity and safety concerns based on their specific properties.

Although there are several different types of 2D nanomaterials, it appears that there is a set of shared characteristics that could be used as a guideline for a standard approach to creating more biocompatible and safer 2D nanomaterials for biomedical applications. Two-dimensional nanomaterial safety information for materials besides graphene is practically nonexistent, and additional evaluations are necessary before promising benefits demonstrated by many 2D nanomaterials can be translated into clinical use [1]. As 2D nanomaterials continue to emerge as viable options in

biomedical applications, it is now time to consider creating an approach for categorizing 2D nanomaterials as “medical grade.”

Overview of Relevant 2D Materials for Biomedical Applications

Graphene Family Nanomaterials

Graphene is a single-atom-thick, two-dimensional sheet having sp^2 -hybridized carbon atoms arranged hexagonally [2]. Its appeal in biomedical applications includes properties, such as enhanced thermal and electrical conductivity, high surface area, and extraordinary mechanical strength [3]. There is an entire set of compounds known as graphene family nanomaterials (GFNs) that use graphene as a base material and include [3]:

- Single/few layer graphene
- Graphene oxide
- Reduced graphene oxide
- Graphene nanoribbons
- Graphene nanosheets

The interesting thing about GFNs, is that even though they all have graphene as a component, they have different physicochemical structure, which affects their toxicity and biocompatibility. Physical variations in properties influence interactions with biological systems, including: purity, lateral dimensions, defect density, layer number, surface area, stiffness,

shape, size, and surface chemistry [3,4]. It has been demonstrated that surface chemistry has the greatest impact on biocompatibility [3]. The two active parts that facilitate attaching to biological molecules and adhering to cells are surfaces and edges [2].

The concentration of graphene is directly related to apoptosis (cell death) [3]. Proteomic analysis shows that graphene exposure causes the expression of 37 proteins, which is still less than the activation in response to single wall carbon nanotubes [3]. The cytotoxicity of graphene may be linked to a reduction in the mitochondrial membrane potential and increment of intracellular reactive oxygen species (ROS) [3]. Graphene has the lowest hemolytic activity of all GFNs, most likely due to the lack of oxygen on its surface and the tendency of it to form aqueous aggregates. These characteristics limit graphene's interactions with red blood cell membranes [3].

Graphene oxide (GO) is the most extensively studied of all GFNs, and has shown the following effects [2,3]:

- Adsorbs to proteins very well
- Damage to plasma membrane due to strong physical interaction with phospholipid bilayer
- Internalized in cells – elevated ROS, altered cellular ultrastructure, diminished metabolic activity
- The smaller the particles, the more hemolysis

The toxicity of GO is due to a combination of oxidative stress and the impairment of plasma membranes because impaired mitochondrial function leads to ROS formation [2,3]. It has been demonstrated that the structure of GO is highly dependent on how it is prepared [3] because highly purified GO produces negligible negative effects in vitro and in vivo, which means that there is a need for considering impurities when attempting to minimize biological effects [2]. In addition, surface charge changes the aggregation status of GO and its ability to be internalized. Lower positive surface charges tend to have milder toxic effects. Therefore, for clinical applications, it will be useful to decrease the electrical surface charge of GO in particular [2].

Reduced graphene oxide (rGO) shares similar characteristics to graphene and GO. Small particles produced 26 times more ROS compared with larger particles, and lateral size is a major factor for toxic effects [3]. Again, surface chemistry plays a vital role in toxicity. In regards to halogenographenes (graphene with halogens - Cl, Br, I - on the surface), chlorographene is the most toxic. Hydrogenated graphene (graphane) appears to be more cytotoxic than graphene oxide. The toxicity of rGO may be linked to the fact that it is less likely to aggregate than graphene oxide [3].

The toxicity of all GFNs appears to be time-, dose-, and size- dependent. A summary of shared characteristics of GFNs that increase their toxicity are [3]:

- Particle size – smaller particles, greater hemolysis
- Oxygen content on surface – more oxygen increases hemolysis
- Method of preparation – Influences defects and amount of oxygen

In addition, GFNs with small size, sharp edges, and rough surfaces internalize into cells more easily than larger, smooth GFNs, and GFNs with more layers appear to lower adsorptive capacity regarding protein binding [2].

One of the key properties of GFNs that makes them so valuable in biomedical applications is their ability to form aggregates, especially in aqueous solutions. Graphene family nanomaterials tend to aggregate in physiological solutions due to electrostatic charges and nonspecific binding to proteins. Functionalizing GFNs, either through covalent conjugation using polymers or noncovalent physisorption, could improve solubility and biocompatibility and reduce cyto- and genotoxicity [2].

Transition Metal Dichalcogenides

Transition metal dichalcogenides (TMDs) are similar to graphene, but are not a single layer like graphene. Instead, they consist of layers of chemically bonded atoms which are attached together and held by Van der Waals' interactions [5]. Their structure is composed of a transition metal, typically molybdenum (Mo), tungsten (W), or Rhenium (Re) and a chalcogen, usually sulfur (S) or selenium (Se). They have the chemical formula MX_2 [M = transition metal; X = chalcogen].

Biomedical applications of TMDs include sensors and possibly tissue engineering; however, there is extremely limited data on these nanomaterials, especially in regards to toxicity. Teo et al. demonstrated that molybdenum disulfide (MoS_2) and tungsten disulfide (WS_2) appear to be less toxic than tungsten diselenide (WSe_2) [6]. These studies have shown that at least these three TMDs may be less toxic than GO [5]. What is interesting is that the preparation method, in this case exfoliation, appears to make a difference regarding toxicity, particularly for MoS_2 . The extent of exfoliation that MoS_2 undergoes is a factor influencing its toxicity. The more exfoliated the MoS_2 nanosheets, the stronger the cytotoxic influence, which may be due to an increase in surface area and active edge sites [5]. Overall, MoS_2 nanoparticles seem to have a very good biocompatibility with human cells [7].

Hexagonal Boron Nitride

Hexagonal-boron nitride (hBN) has a hexagonal structure like graphene, but is composed of alternating boron (B) and nitrogen (N) atoms arranged in a monolayer. This structure is less electronegative than carbon. It has low density, high thermal conductivity, chemical stability, and easy workability [8]. Although little is known about the toxicity of hBN, boron itself has low acute toxicity [8], and hBN may have better biocompatibility than graphene [1]. Recently, Atila et al. implanted a composite of hBN and hyaluronic acid (HA) h-BN nanoparticles in rat femurs [8]. While formal toxicological assays were not conducted, no increase of serum B levels was detected demonstrating that the hBN did not degrade [8].

Other 2D Nanomaterials

In the burgeoning field of 2D nanomaterial development, other possibilities for biomedical applications include polymeric nanosheets, silicene, germanene, layered double hydroxides, phosphorene, and stanene. However, no toxicological data exists for any of these nanomaterials, and standard methodologies for their fabrication must be developed.

Developing a Standard Model

There is an immediate need to focus on identifying a library of 2D nanomaterials that are appropriate for various biomedical applications [1]. A strong prerequisite of any community approaching the study of nanomaterials toxicity is sharing a classification approach for the nanomaterial itself [9]. Utilizing the shared findings among toxicity data available for GFNs, TMDs, and hBN, and assuming that some characteristics would apply overall to the group of 2D nanomaterials, it could be assumed that medical grade 2D nanomaterials would have the following characteristics, with no particular priority:

- More than one layer – an optimal number would depend on the specific nanomaterial being used and its biomedical function
- Standard preparation by a method suitable for the intended nanomaterial that would minimize the amount of impurities
- A low surface oxygen content
- A weak, positive surface charge
- Particle size as large as possible to minimize toxicity but is still optimal for functionality
- Particles that aggregate, especially in aqueous solution
- Particles with smoother surfaces that have minimal protein adsorption potential but retain functionality

Proper assessment of safety profiles and impact on health and the environment is a fundamental prerequisite to translate any development

into action from industrial applications to healthcare [9]. Using the limited toxicological data available on 2D nanomaterials to this point, a hierarchy of preferred 2D nanomaterials for biomedical use can be established. From greatest biocompatibility to least, the preference in order for biomedical applications would be:

1. Hexagonal boron nitride
2. Graphene
3. Molybdenum disulfide/tungsten disulfide
4. Tungsten diselenide
5. Graphene oxide
6. Reduced graphene oxide

This schema fits into the four fundamental steps needed to achieve significant results in the assessment of a safety profile of graphene and 2D materials as outlined by Bianco et al. [9]. As stated by Nezakati, biocompatibility and toxicity behavior of graphene-based materials (and 2D nanomaterials in general) in biological systems gives rise to many important fundamental issues that require significant attention [10]. Oxidative stress may be the result of immune responses elicited by materials or properties of materials themselves. Some 2D nanomaterials also contain metals not usually found above trace levels in humans [1]. Rational material design will remain an important and leading approach of the community, but strategies that are complemented by biological assays and analyses will allow mining of huge datasets to evolve new hypotheses for improved biomaterial design [11].

Conclusion

The recent explosion of 2D nanomaterial development has provided a need to identify better methods of standardization, not only for the materials themselves, but also risk management, testing methods, and characterization [10]. One major area lacking is a system for defining which types of 2D nanomaterials would be safe and effective enough for biomedical applications. The extremely limited toxicology data is challenging, but the available literature does show trends among the 2D nanomaterial types that could be integrated into a usable model, at least currently. As new data are collected, this model can be expanded to include new information and adjusted accordingly. The approach presented here can be used as a model to at least begin the arduous task of unifying the

field of 2D nanomaterials for medical use.

References

1. Chimene D, Alge DL, Gaharwar AK (2015) Two-Dimensional Nanomaterials for Biomedical Applications: Emerging Trends and Future Prospects. *Adv Mater* 27: 7261-7284.
2. Guo X, Mei N (2014) Assessment of the toxic potential of graphene family nanomaterials. *J Food Drug Anal* 22: 105-115.
3. Chng E, Pumera M (2015) Toxicity of Graphene Related Materials and Transition Metal Dichalcogenides. *RSC Adv* 5: 3074-3080.
4. Sanchez VC, Jachak A, Hurt RH, Kane AB (2012) Biological interactions of graphene-family nanomaterials: an interdisciplinary review. *Chem Res Toxicol* 25: 15-34.
5. Chng EL, Sofer Z, Pumera M (2014) MoS₂, exhibits stronger toxicity with increased exfoliation. *Nanoscale* 6: 14412-14418.
6. Teo WZ, Chng EL, Sofer Z, Pumera M (2014) Cytotoxicity of exfoliated transition-metal dichalcogenides (MoS₂, WS₂, and WSe₂) is lower than that of graphene and its analogues. *Chemistry* 20: 9627-9632.
7. Wu H, Yang R, Song B, Han Q, Li J, et al. (2011) Biocompatible Inorganic Fullerene-Like Molybdenum Disulfide Nanoparticles Produced by Pulsed Laser Ablation in Water. *ACS Nano* 5: 1276-1281.
8. Atila A, Halici Z, Cadirci E, Karakus E, Palabiyik S, et al. (2016) Study of the Boron Levels in Serum after Implantation of Different Ratios Nano-Hexagonal Boron Nitride-Hydroxy Apatite in Rat Femurs. *J Mat SciEng C* 58: 1082-1089.
9. Bianco A, Prato M (2015) Safety Concerns on Graphene and 2D Materials: A Flagship Perspective. *2D Mater* 2: 1-3.
10. Nezakati T, Cousins BG, Seifalian AM (2014) Toxicology of chemically modified graphene-based materials for medical application. *Arch Toxicol* 88: 1987-2012.
11. Tibbitt MW, Rodell CB, Burdick JA, Anseth KS (2015) Progress in material design for biomedical applications. *Proc Natl Acad Sci USA* 112: 14444-14451.